effect intermediate to that of cAMP. However, in the C-helix cAMP and cCMP exhibit similar effects, but cGMP produces an intermediate effect between the apo and bound cAMP/cCMP DEER distance distributions. These data provide an interesting lens for studying allostery in HCN channels and indicate that the mechanism of protein allostery in the CNBD varies for different cyclic nucleotides.

968-Plat
Rapid Activation of Distinct Conducting States in P2X Receptor Channels
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P2X receptors are trimeric cation channels that are activated by extracellular ATP. These purinergic receptors have been reported to undergo pore dilation following activation by high concentrations of ATP, a phenomenon thought to mediate apoptosis in the immune system. Here we demonstrate that the widely reported slow time-dependent increase in the relative permeability of NMDG to Na, thought to reflect pore dilation, results from a gradual inhibition of small-cation-selective channels following depletion of intracellular alkali ions. Moreover, we find that P2X receptors enter both small-cation-selective open states and large-cation-permeable open states within milliseconds of ATP application. Taken together, our results demonstrate that P2X receptors can rapidly enter a large-cation-permeable open state without requiring either high ATP concentrations or high channel density, indicating that the ability of ATP to permeabilize cells can occur under physiologically realistic conditions.

Platform: Intrinsically Disordered Proteins (IDP)

969-Plat
A New and Un-Conventional Ultrafast Binding Mechanism of Intrinsically Disordered Proteins to Structured Partners
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Intrinsically disordered proteins (IDPs) mediate several and very diverse processes such as signaling, regulation and formation of entropic barriers. Such functions often involve the binding between an IDP and one or more structured partners. Several mechanisms to describe the binding of IDPs to structured proteins have been identified: the *conformational selection* model implies that, along the conformational ensemble explored by the IDP, binding-prone configurations are abundant and favor binding. In contrast, the *induced-fit* model describes binding as a series of conformational changes of the IDP that can form secondary structure elements upon binding. By investigating the association between the intrinsically disordered nucleoporin Nup153 and its structured binding partner Importin-β through means of single-molecule FRET experiments, molecular dynamics (MD) and Brownian dynamics (BD) simulations, we have identified a new mechanism that leads to the formation of complexes without the need of any structural rearrangement or selection of a pre-configured conformation of the IDP. We found that binding can occur between Importin-β and highly diverse configurations of Nup153 in a globular-like state. Being conformational-independent, the association is solely regulated by the availability of nucleoporin's Phe-Gly dipeptide responsible for the interaction with Importin. More importantly, since conformational adaptations are absent, specific binding is rapid enough to be observed on the sub-microsecond time scale of the MD simulations. This ultrafast binding mechanism of nucleoporin to Importin-β can justify the observation of rapid, yet selective nucleo-cytoplasmatic transport. Our joint computational and experimental approach could help to explain if a similar binding scenario also applies to other repetitive IDPs.

970-Plat
Combinational Evidence that Intrinsic Disorder Provides Broad Association Profiles
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Using a simple lattice model approach that captures the combinatorial essence of a short intrinsically disordered peptide binding to a larger protein surface we provide evidence that non-specific peptides with moderate amounts of disorder can bind to a larger array of protein surfaces than their ordered counterparts.

971-Plat
The Intrinsically Disordered C. Trachomatis Tarp Binds Actin with a Partially Preformed Helix
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Tarp (Translocated actin recruiting protein) is an intrinsically disordered protein (IDP) from *Chlamydia trachomatis*, which is translocated into the host cell via type III secretion to initiate the infection process. Once exposed to the cytosol, Tarp recruits host-cell actin, forming actin pedestal-like structures that contribute to host cell invasion (Clifton *et al.*, *PNAS***101**,10166). Tarp is capable of directly nucleating the formation of actin filaments, and its actin-binding region was mapped to the region between residues 726 and 825 (Jewett *et al.*, *PNAS***103**, 15599). We have acquired and assigned nuclear magnetic resonance (NMR) spectra of $Tarp_{726\text{-}825}$, which show the narrow spectral distribution and chemical shifts characteristic of an IDP. Tarp shows some homology to the actin binding WH2 motif from WAVE2, and therefore it is expected to bind in a similar manner, forming a helix upon binding. Indeed, NMR relaxation experiments of $Tarp_{726\text{-}825}$ show slightly lower flexibility in the 10-residue region homologous to the WH2 motif. However, chemical shift index calculations show that this helix is only partially formed before binding. In the presence of globular actin, the majority of peaks in the Tarp NMR spectra are affected, indicating that the binding affects most of the amino acid residues in this Tarp fragment. Isothermal titration calorimetry experiments determined the dissociation constant for the binding to be $1.02\ (\pm\ 0.32) \times 10^{-7}$ M, and showed a significant decrease in entropy upon binding, indicating increased order upon binding. Based on this information, the binding of Tarp to actin was modelled *in silico.* Two possible low energy sites of interaction have been identified, both of which could account for these observations. It is therefore possible that Tarp binds actin through a ''fuzzy'' interface, with contribution from both conformations.

972-Plat
Post-Translational Modification of P27 Regulates Signal Transmission via a Dynamic Interaction with Cdk2/Cyclin
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Proteins that are fully or partially unstructured are named intrinsically disordered proteins (IDP). The function of IDP's, in context of the structure-function paradigm, is challenged by their inherent flexibility. The current hypothesis of induced-folding mechanism of IDP's suggests many advantages over well-ordered and structured proteins because of it ability to recognize and regulate multiple pathways by adopting numerous conformations. One of the most prominent examples is the disordered polypeptide p27. p27 is a small IDP that inhibits cyclin-dependent kinases (Cdk) regulating cell proliferation. Furthermore, phosphorylation of p27 leads to its degradation by the ubiquitin-protease pathway. By applying single-molecule fluorescence spectroscopy in combination with biochemical assays, we tested the hypothesis that bound disordered regions of p27 are essential for its regulatory functions. However, the existence of regulatory modification sites within bound, folded regions of disordered proteins raises a key question regarding how these sites become accessible for enzymatic modification. Our results show that p27 is dynamic even when bound to the Cdk2/CyclinA complex. Moreover, sequential phosphorylation at positions Y88 and Y74 of p27 relieves Cdk2 inhibition with rheostat-like precision. These observations explain the mechanism of how intra and inter-molecular phosphorylation of partially activated Cdk2/CyclinA are regulated during the cell cycle and provide the basis for understanding how dynamics within Cdk2/CyclinA-bound p27 mediate signal transmission.

973-Plat
Effects of Charge Interactions and Transient Secondary Structure Elements on the Function of the Disordered RAM Region of the Notch Receptor
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The Notch signaling pathway is crucial for cellular development. The intracellular domain of the Notch receptor (NICD) contains the 140-residue intrinsically disordered RAM region followed by seven ankyrin repeats, a nuclear localization sequence, and a C-terminal degradation sequence. The RAM